ISSN:0975-3583,0976-2833 VOL12,ISSUE 05,2021

Diagnostic and Prognostic Value of Ascitic Prostaglandin E2 in Cirrhotic Patients with Spontaneous Bacterial Peritonitis

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Abstract

Background: Spontaneous bacterial peritonitis (SBP) is frequent in cirrhosis and represents the most common cause of hospitalization. Despite progress in their pathogenesis, prevention and management, bacterial infections still remain a cause of mortality and admission to intensive care units. This study is aimed to detect the role of ProstaglandinE2 (PGDE2) in serum and ascetic fluid as a diagnostic marker for eradication of SBP. **Patients and methods:** A prospective cohort study involved patients with liver cirrhosis, ascites and spontaneous bacterial peritonitis referred to the Internal Medicine, Faculty of Medicine, Zagazig University. Patients with ascites divided into 2 groups: case group (26 cirrhotic patients with SBP) ,control group (26 cirrhotic patients without SBP). All patients were subjected to a thorough history and complete clinical examination. White blood cell and Polymorphonuclear cells (PMN) counts in peripheral blood; PMN counts, protein, glucose in ascitic fluid and PGDE2 before and after five days of treatment were estimated. **Results:** Serum and ascitic PGDE2 was elevated in all cirrhotic groups either case and control more than normal. However, PGDE2 level was lower in case group before treatment in comparsion with control group, and after treatment PGDE2 levels was elevated. There is statistically significant change in PMN after treatment**.** The best cutoff of ascitic fluid PGDE2 in diagnosis of SBP among ascitic patients is ≤ 750.512 with area under curve 0.71, sensitivity 76.9%, specificity 61.5% , positive predictive value 66.7%, negative predictive value 72.7%, accuracy 69.2% ($p<0.05$). Regarding performance of serum PGDE2, ROC curve showing area under curve 0.781, sensitivity 76.9%, specificity 76.9%**.** There is statistically significant positive correlation between ascitic fluid prostaglandin E2 before treatment and serum prostaglandin E2 before treatment. **Conclusion:** Prostaglandin E2 as acute phase reactant increased in patient with SBP. Serum or ascitic ProstaglandinE2 was high in SBP and declined in patients responding to antibiotic treatment. PMN counts in peripheral blood didn't decrease in response to antibiotic treatment of SBP.

Keywords: Spontaneous Bacterial Peritonitis PGDE2 ; PMN; Cirrhosis

Introduction:

Spontaneous bacterial peritonitis (SBP) is the progress of a bacterial infection in the peritoneum causing peritonitis, in spite of the absence of a noticeable cause for the infection. It occurs almost completely in people with portal hypertension, usually due to cirrhosis of the liver **(1).** Structural and functional changes in the intestinal mucosa increase its permeability and result in Bacterial translocation. The intestinal barrier includes secretory and natural defense mechanisms against microorganisms. Intestinal mucosa and intercellular junctions among epithelial cells form a layer that allows selective passage of the toxins and bacterial products **(2).**

Patients with cirrhosis who are in a decompensated state are at the highest risk of developing spontaneous bacterial peritonitis. Low complement levels are associated with the development of spontaneous bacterial peritonitis **(3)**. Patients with low protein levels in ascitic fluid (< 1 g/dL) have a 10-fold higher risk of developing spontaneous bacterial peritonitis than those with a protein level greater than 1 g/dL and elevated serum bilirubin level and serum albumin level less than 2.85 g/dL **(4)**.

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The particular susceptibility of patients with cirrhosis to infections is related to an immunodeficient state due to the concomitant presence of various facilitating mechanisms. In cirrhosis there are changes in the intestinal flora and intestinal barrier, reduced reticuloendothelial function, deficiencies in C3 and C4, decreased opsonic activity of the ascitic fluid and neutrophils leukocyte dysfunction **(5)**.

It is well known that Prostaglandin E2 (PGE2) has a variety of immunosuppressive functions including inhibition of macrophages phagocytosis and killing activity, neutrophils chemotaxis and production of proinflammatory mediators in leukocytes, thus resulting in the down regulation of immune functions and impairing host defense against microorganisms **(6)**. It was found that PGE2 plays a key role in the development of immune paralysis by suppressing proinflammatory cytokine secretion and bacterial killing of macrophages in patients with end-stage liver disease and acutely decompensated cirrhosis **(7).**

The aim of the present study was to detect the role of ProstaglandinE2 (PGDE2) in serum and ascetic fluid as a prognostic marker for eradication of SBP.

Patients and methods:

A prospective cohort study involved 52 patients with liver cirrhosis and ascites referred to the Internal Medicine, Faculty of Medicine, Zagazig University, during the period from June 2020 to March 2021. Patients with ascites divided into 2 groups: case group (26 cirrhotic patients with SBP),control group (26 cirrhotic patients without SBP). Written consents were taken from all patients according to declaration of Helsinki.

Inclusion criteria:

Decompensated cirrhotic patients with ascites and diagnosed as SBP of both gender; Male and female.

Exclusion criteria

Patient with Collagen vascular disorders, patient with ascites who did not have the criteria of sbp, patient with any form of acute arthritis, patient with acute infections and septicemia and patient with other causes of elevated PGDE2

Diagnosis of liver cirrhosis:

It was done by clinical signs, laboratory and ultrasound findings and severity of the liver disease was scored according to Child–Pugh's classification. Child-Pugh A=5-6 points, Child-Pugh B=7-9 points, Child-Pugh C=10 or more points **(8).**

All patients were subjected to a thorough history, complete clinical examination and routine investigations including :Complete blood picture (by automated blood counter); Liver function tests: serum bilirubin (total and direct), serumalbumin, serum ALT and AST measured by kinetic method; Renal function tests: serum creatinine, urea; Coagulation profile: PT, PTT and INR.

Radiology investigations:

Ultrasonography for diagnosis of cirrhosis as a shrunken liver (smalland nodular), enlarged spleen (splenomegaly) and portal hypertension (dilated portal vein, hilar varices), Ascites, HCC

Special Investigations:

PGDE2 measurement in serum&ascitic fluid and ascites PMNs measurement. The PGDE2 ELISA is based on the principle of a solid phase enzyme-linked immunosorbent assay. The assay system utilizes a unique monoclonal antibody directed against a distinct antigenic determinant on the on the PGDE2molecule.

Follow up:

After 5 days from starting treatment of SBP by ceftriaxone 2g/24h or cefotaxime 2g/8h: 3 cm blood was taken: for measuring serum PGDE2. Ascitic fluid sample containing 5 cm by paracentesis with all aseptic precautions for
PMN and ascitic fluid PGDE2.

Statistical analysis:

ISSN:0975-3583,0976-2833 VOL12,ISSUE 05,2021

All data were analyzed using SPSS 20.0 for windows (SPSS Inc., Chicago, IL, USA) and MedCalc 13 for windows (MedCalc Software bvba, Ostend, Belgium). ANOVA test, Independent Student t-test, Mann-Whitney U, Chi-square test or Fisher's exact test and Kraskall Wallis H were used.. Post hoc test for multiple comparisons was done by using LSD or Tamhane's T2 method according to homogeneity of the variance. Spearman's rank correlation coefficient was calculated to assess correlations between study parameters. We consider (+) sign as indication for direct correlation & (-) sign as indication for inverse correleation. Receiver operating characteristic (ROC) curve analysis was used and area Under Curve (AUROC) was calculated as follows: 0.90:1= excellent, 0.80:0.90 = good, 0.70:0.80 = fair; 0.60:0.70 = poor; and 0.50:0.6 = fail. The optimal cutoff point was established at point of maximum.

RESULTS

The present study showed a significant difference between the studied groups regarding serum and ascitic fluid prostaglandin E2 which were higher among control group **(Figure 1)**. Serum and ascitic PGDE2 was elevated in all cirrhotic groups either case and control more than normal. However, PGDE2 level was lower in case group before treatment in comparsion with control group, and after treatment PGDE2 levels was elevated **(Table 1).**

The best cutoff of ascitic fluid PGDE2 in diagnosis of SBP among ascitic patients is ≤ 750.512 with area under curve 0.71, sensitivity 76.9%, specificity 61.5% , positive predictive value 66.7%, negative predictive value 72.7%, accuracy 69.2% ($p<0.05$) **(Table 2)**. Regarding performance of serum PGDE2, ROC curve showing area under curve 0.781, sensitivity 76.9%, and specificity 76.9% **(Figure 2).**

There is statistically significant positive correlation between ascitic fluid prostaglandin E2 before treatment and serum prostaglandin E2 before treatment. On the other hand, there is non-significant correlation between AF PGDE2 and age or other laboratory parameters. There is statistically significant correlation between serum prostaglandin E2 before treatment and all of serum albumin , bilirubin, ascitic fluid PGDE2 before treatment. There is statistically non-significant correlation between serum prostaglandin E2 before treatment and age or other laboratory parameters **(Table 3)**.

There is non-significant relation between ascitic fluid PGDE and either child pough class, encephalopathy, grades of ascites or HCC **(Table 4)**. There is non-significant relation between serum PGDE and either grade of ascites, encephalopathy or HCC. There is significant relation between serum PGDE and child pough class with the difference is significant between child B and C **(Table 5)**. There is non-significant relation between mortality and ascitic fluid and serum PGDE2 before and after treatment among case group **(Table 6)**.

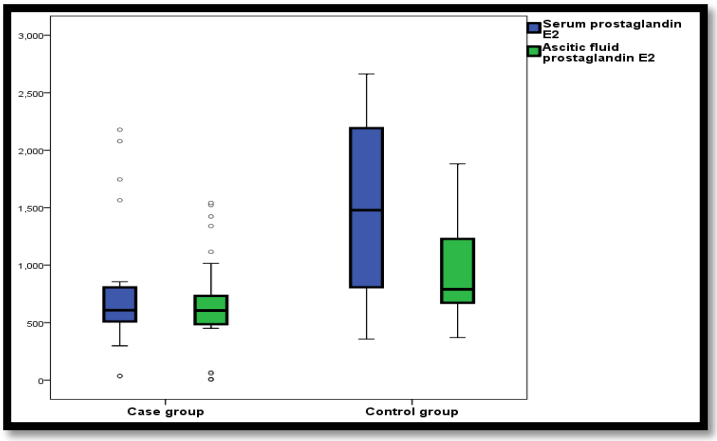

Figure (1) Boxplot showing comparison between the studied groups regarding serum and ascutuc fluid PGDE2

ISSN:0975-3583,0976-2833 VOL12,ISSUE 05,2021

Table (1) comparison between ascitic, serum PGDE2 and ascitic PMN among case group before and after treatment:

	Before	After	Wx	p
	Median (range)	Median (range)		
Ascites PGDE2	605.38 (4.55 – 1541.92)	903.79 (502.28 – 1760)	-2.476	0.013*
Serum PGDE2	608.25 (34.15 – 2178.94)	1134.72 (527.79 – 1769.98)	-1.944	0.049*
Ascites PMN	545 (270 – 5600)	210 (160 – 240)	-4.459	<0.001**

Wx Wilcoxon signed rank test *p<0.05 is statistically significant **p≤0.001 is statistically highly significant

Table (2) Performance of ascitic fluid PGDE2 in diagnosis of SBP among patients with ascites:

Cutoff	AUC	sensitivity	specificity	PPV	NPV	Accuracy	P
≤750.512	0.71	76.9%	61.5%	66.7%	72.7%	69.2%	0.009*

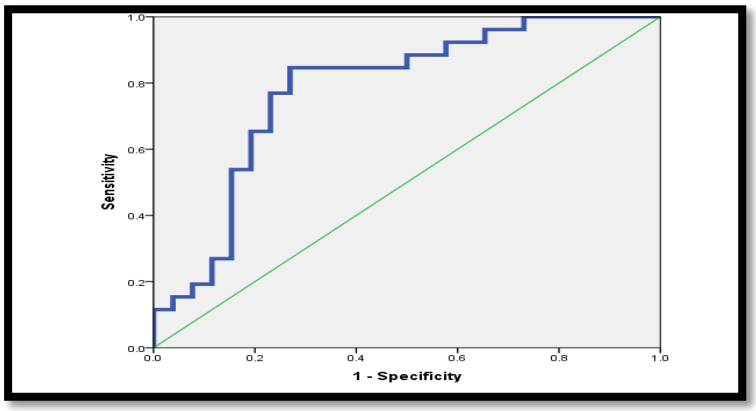

Figure (2) ROC curve showing performance of serum PGDE2 in diagnosis of SBP among patients with ascites

Table (3) Correlation between serum, ascitic fluid PGDE2 before treatment and both age and laboratory parameters among the studied patients:

ISSN:0975-3583,0976-2833 VOL12,ISSUE 05,2021

	Ascitic fluid PGDE2		Serum PGDE2	
	r	p	r	p
Age (year)	-0.075	0.597	-0.027	0.849
Hemoglobin	0.233	0.096	0.172	0.224
TLC	-0.101	0.474	-0.113	0.425
Platelet count	0.157	0.266	0.087	0.539
S. bilirubin	-0.156	0.271	-0.378	0.006*
S. albumin	0.232	0.097	0.422	0.002*
ALT	0.172	0.222	0.015	0.916
s. creatinine	0.024	0.865	-0.034	0.809
PT	-0.025	0.861	-0.224	0.11
INR	0.083	0.557	-0.075	0.596
AF PMN	0.009	0.966	-0.091	0.657
Serum PGDE2	0.535	<0.001**		

r Spearman rank correlation coefficientWx Wilcoxon signed rank test *p<0.05 is statistically significant **p≤0.001 is statistically highly significant

Table (4) Relation between ascitic fluid PGDE2 before treatment and present history of the studied patients:

	Ascitic fluid PGDE2		Test	
	Median	Range	KW/Z	p
Child pough				
B	887.32	4.55, 1882.03	5.255	0.072
C	657.22	8.18, 1541.92		
Encephalopathy:				
No	697.06	4.55, 1882.03	-0.152	0.849
Yes	655.44	369.52, 1340.82		
HCC:				
No	691.59	4.55, 1882.03	0.308	0.758
Yes	703.08	8.68, 1541.92		
Ascites:				
Moderate	768.07	450.03, 1882.03		
Tense	673.06	4.55, 1780.01	-1.044	0.297

KW Kruskal Wallis test Z Mann Whitney test

Table (5) Relation between serum PGDE2 before treatment and present history of the studied patients:

	Median	Range	KW/Z	p
Child pough				
B	1696.14[2,3]	597.67, 2556.19	12.355	0.002*
C	687.86[2,3]	34.15, 2662.99		
Encephalopathy:				
No	848.62	34.15, 2662.99	-1.293	0.205
Yes	612.53	357.05, 2192.52		
HCC:				
No	906.11	298.53, 2556.19	-1.116	0.265
Yes	746.25	34.15, 2662.99		
Ascites:				
Moderate	1056.19	366.69, 2662.99		
Tense	800.51	34.15, 2556.19	-1.024	0.306

KW Kruskal Wallis test Z Mann Whitney test

ISSN:0975-3583,0976-2833 VOL12,ISSUE 05,2021

Table (6) Relation between serum, ascitic fluid PGDE2 and mortality among the studied patients:

	Ascites fluid Median (range)	p	Serum Median (range)	p
	Before treatment			
Mortality:				
No	574.94(4.55 – 1541.92)	**0.123**	762.96 (38.12 –2178.94)	0.394
Yes	1078.57(73.96 – 1541.92)		370.47 (34.15 – 706.79)	
	After treatment			
Mortality:				
No	853.44 (02.28 – 1760)	0.185	697.68 (390.02 – 1092.18)	0.258
Yes	1145.35 (1086.15-1204.56)		860.69 (719.19 – 1002.15)	

P for Mann Whitney test

Discussion:

Spontaneous bacterial peritonitis (SBP) is the most common infectionresponsible for sepsis-induced acute-on top of chronic liver disease **(9)**.

In patients with SBP, the mortality rate may be as high as 30% in hospital despite good infection control measures, mortality being generally due to complications such as acute variceal bleeding, hepatorenal syndrome or advanced liver failure **(10)**. The diagnosis of SBP is based on a PMN leukocyte count (PMN >250/mm3) in ascetic fluid. The diagnosis of SBP (based on a PMN >250/mm3) does not take into account bacteri-ascites, which is a variant of SBP where a single bacterial organism grows in ascitic fluid but the number of PMN is <250/mm3 **(9)**.

Prostaglandin E2 (PGE2) reflects the degree of systemic inflammation, regardless of the underlying cause. Patients with decompensated cirrhosis.PGD E2 as a surrogate marker of systemic inflammatory response and PGD E2 levels were higher in patients who developed systemic inflammatory response syndrome, infection, and alcoholic hepatitis **(11).**

This clinical-based prospective cohort study involved 52 cirrhotic patients with ascites divided into 2 groups: case group (26 cirrhotic patients with SBP) ,control group (26 cirrhotic patients without SBP) admitted at the Internal Medicine hospital, Faculty of Medicine, Zagazig University Hospitals. The aim of this study was to establish the role of PGDE2 in serum and ascitic fluid asmarker for diagnosis and detection of SBP.

Our results showed serum and ascitic PGDE2 was elevated in all cirrhotic groups either case and control more than normal. However, PGDE2 level was lower in case group before treatment in comparsion with control group, and after treatment PGDE2 levels was elevated. Our study was compared with a previous studies; **Rizk et al. (12)** who concluded that serum PGDE2 correlates well with the PMN count and protein levels in ascitic fluid and reliably diagnoses. **Luo et al. (13)** who studied role of ascitic PGDE2 in diagnosis of spontaneous bacterial peritonitis and prediction of in-hospital mortality in patients with decompensated cirrhosis SBP. We found that ascitic PGE2 levels in patients with SBP decreased significantly, which may serve as a biomarker of indicating SBP (the cut-off value was 40.3 pg/mL). An AUC of 0.75 suggested that ascetic PGE2 was an intermediate biomarker for diagnosis of SBP. Ascitic PGE2 was a better biomarker indicating SBP than serum CRP, and not superior to WBC, neutrophils, and serum PCT. Thus, the diagnostic value of ascitic PGE2 was not higher than that of WBC, neutrophils, or serum PCT, and it needed to be combined with other inflammatory markers, such as WBC, while, the measures of WBC and neutrophils in clinical were faster and easy-to-be-detected.

It is well known that patients with SBP have a much worse immunity status, and present with impaired function and reduced numbers of macrophages in ascites **(14;15)**. Thus, the PGE2 in ascites was lower than patients with cirrhosis without spontaneous bacterial peritonitis.

ISSN:0975-3583,0976-2833 VOL12,ISSUE 05,2021

In Weiler's study, PGE2 tissue levels were significantly decreased in inflamed gastric antral mucosa of patients with cirrhosis in the presence of portal hypertension, and further decreased in inflamed gastric antral mucosa of patients with ulcers **(16).** While **Shahed and Shoskes (17)** observed a higher level of PGE2 in prostatic secretions of men with symptomatic chronic prostatitis.

Actually no data were disclosed on the comparison of the ascitic PGE2 concentration in the decompensated cirrhotic patients with or without SBP.

This study had some strength points. The first that good randomizations led to statistically non-significant difference between serum PGDE2 levels and ascitic fluid PMN concerning study parameters making drug used is the only responsible factor for any change and removing possible confounders. The second that the nature of the study as prospective ones excluding any possible recording or recalling bias.

Yet, it had some limitations; it is performed in a single center and relatively small sample size. We recommend further large scale multicentric prospective studies to validate our findings.

Conclusion:

Prostaglandin E2 as acute phase reactant increased in patient with SBP. Serum or ascitic Prostaglandin E2 was higher in patients responding to antibiotic treatment. PMN counts in peripheral blood didn't decrease in response to antibiotic treatment of SBP.

No conflict of interest.

REFERENCES:

1- **Sheikhbahaei, S., Abdollahi, A., Hafezi-Nejad, N., Zare, E. (2014)**. Patterns of antimicrobial resistance in the causative organisms of spontaneous bacterial peritonitis: a single centre, six-year experience of 1981 samples. International journal of hepatology, 2014.

2- Bellot P, García-Pagán JC, Francés R, et al.(2010): Bacterial DNAtranslocation is associated with systemic circulatory abnormalities and intrahepatic endothelial dysfunction in patients with cirrhosis. Hepatology;52:2044-2052.

3- **Runyon BA, and AASLD.(2015):** Introduction to the revised AmericanAssociationfor the Study of Liver Diseases Practice Guideline managementof adult patients with ascites due to cirrhosis 2012. Hepatology. Apr. 57 (4):1651-3.

4- **Huang CH, Lin CY, Sheen I.(2011):** Recurrence of spontaneousbacterial peritonitis in cirrhotic patients non-prophylactically treated with norfloxacin: serum albumin as an easy but reliable predictive factor. Liver Int; 31: 184–91.

5- **Giulia Pieria, Banwari Agarwalb, Andrew K. et al. (2014).** "C-reactive protein and bacterial infection in cirrhosis".Ann Gastroenterol; 27 (2): 113-120.

6- **Bordon AP, Dias-Melicio LA, Acorci MJ, Calvi SA, SerraoPeracoli MT, Victoriano AM (2007).** Prostaglandin E2 inhibits Paracoccidioides brasiliensis killing by human monocytes. Microbes Infect; 9(6): 744-747**.**

7- **O'Brien AJ, Fullerton JN, Massey KA, Auld G, Sewell G, James S, et al(2014).** Immunosuppression in acutely decompensated cirrhosis is mediated by prostaglandin E2. Nat Med; 20(5): 518-523.

8- **Kim, H. J., & Lee, H. W. (2013).** Important predictor of mortality in patients with end-stage liver disease. Clinical and molecular hepatology, 19(2), 105.

9- **Fiore, M., Maraolo, A. E., Leone, S., Gentile, I., Cuomo, A., Schiavone, V., Cascella, M. (2017)**. Spontaneous peritonitis in critically ill cirrhotic patients: a diagnostic algorithm for clinicians and future perspectives. Therapeutics and clinical risk management, 13, 1409.

10- **Ghassemi, S., & Garcia-Tsao, G. (2007).** Prevention and treatment of infections in patients with cirrhosis. Best Practice & Research Clinical Gastroenterology, 21(1), 77-93.

11- **Cervoni JP, Thevenot T, Weil D, et al.(2012):** C-reactive proteinpredicts short-term mortality in patients with cirrhosis. J Hepatol; 6:1299-1304.

12- **Rizk E, Elzehery R, Zakaria S. (2014).** Ascitic fluid calprotectinand serum C-reactive protein as diagnostic markers for spontaneous bacterial peritonitis. Afro-Egypt J Infect Endem Dis; 4(3): 117-125.

ISSN:0975-3583,0976-2833 VOL12,ISSUE 05,2021

13- **Luo, J., Wu, X., Zhang, Y., Huang, W., & Jia, B. (2019).** Role of ascitic prostaglandin E2 in diagnosis of spontaneous bacterial peritonitis and prediction of in-hospital mortality in patients with decompensated cirrhosis. Medicine, 98(26).

14- **Nieto JC, Sanchez E, Romero C. (2015):** Impaired innate immune response of leukocytes from asciticfluid of patients with spontaneous bacterial peritonitis. J Leukocyte Biol;98:819–25.

15- **Fagan KJ, Rogers GB, Melino M, et al**. **(2015):** Ascites bacterial burden and immune cell profile are associated with poor clinical outcomes in the absence of overt infection. PLoS One 2015;10:e0120642.

16- **Weiler H, Weiler C, Gerok W. (1990):** Gastric mucosal prostaglandin E2 levels in cirrhosis and portal hypertension. J Hepatol 1990;11:58–64.

17- **Shahed AR, Shoskes DA.(2001):** Correlation of beta-endorphin and prostaglandin E2 levels in prostaticfluid of patients with chronic prostatitis with diagnosis and treatment response. J Urol;166:1738–41.